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THE DETERMINATION OF AMMONIA IN SOILS

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THE DETERMINATION OF AMMONIA IN SOILS

By R. S. Potter and R. S. Snyder

INTRODUCTION.

Theoretically, the ideal method for determining ammonia in soils would give the absolute amount of ammonia present as such and as the ammonium radical, but in the light of present knowledge, it cannot be said beyond all doubt that any conceivable method would give this ideal result. This is true because a large part of the nitrogen of the soil is present in protein and protein degradation products, the extensive decomposition of which gives large amounts of ammonia. The uncertainty regarding just what products are present does not permit the finding of conditions which we can be certain will expel ammonia already present and not decompose any material to give, among other products, ammonia. The problem is further complicated by the well known absorptive and adsorptive powers of the soil.

REQUIREMENTS OF A METHOD FOR AMMONIA.

Therefore, since this ideal result is not attainable, in our opinion the value of any method will depend upon whether it fulfills the following requirements:

1st. Closely agreeing duplicate results should be given and the same result obtained whether the reagent or reagents act, within reasonable limits, for a longer or shorter period. Of course, the reagent or reagents must not include any which are known to decompose material contained in the soil to give ammonia. For instance, it would not be permissible to use strong hot hydrochloric acid, for this reagent decomposes proteins yielding large amounts of ammonia.

2nd. Upon the addition of a known amount of ammonia, the method must give this added amount plus that previously found in the soil. Not enough time, of course, should elapse between the addition of the ammonia and the determination for any bacterial action to take place. In soils already containing ammonia and, apparently all soils do, it is difficult to see how any chemical reaction could, in a reasonably short length of time, change the added nitrogen to some other combination than the ammonium. Because his method does not recover all added ammonia, Russel assumes the ammonia to be chemicaly changed. This will be further discussed later.

3rd. For use in a soils laboratory, the method should

permit one to run several determinations within a reasonable length of time.

METHODS WHICH HAVE BEEN PROPOSED.

The folloing methods have been proposed:

I. Schloesing's Method[1] "A"
II. Schloesing's Method[1] "B"
III. Boussingault's Method[2]
IV. Wolf & Knop's Method[3]
V. Baumann's Method[4]
VI. Russel's Method[5] "A"
VII. Russel's Method[5] "B"

I. Schloesing's first method consists in leaving a mixture of the soil with strong sodium hydroxide solution under a bell jar, together with a vessel containing some standard acid to absorb the ammonia evolved. This method has fallen into complete disuse because of the recognition of the fact that strong sodium hydroxide solution, even in the cold, would graduall decompose the organic nitrogen compounds to give ammonia.

II. In Schloesing's second method, the soil is treated with dilute hydrochloric acid until the liquid remains distinctly acid. The mixture is then well shaken, filtered and an aliquot portion is distilled with alkali. Since this method complies with the third requirement named above, and since we can find no evidences in the literature that it does or does not comply with the first two, we have given the method a rather detailed examination, the results of which are reported and discussed in the experimental part of this paper.

III. In Boussingault's method a mixture of about one part of soil to two parts of water, together with a little magnesia, is distilled into standard acid. It is quite generally admitted that this method does not give true results with material like soil which contains protein and protein degradation products. Yet, because of its ease and simplicity of manipulation, it is often used, it being stated that the results, although not absolute, are comparable with one another. This method has also been examined by us and the results will be taken up in the experimental part of this paper.

IV. The method of Wolf & Knop consists in treating the soil with sodium hypobromite, which reacts with the ammonia, liberating nitrogen gas, which is measured. Baumann[4] has shown this method does not give accurate results.

V. In Baumann's method a hydrochloric acid extract is made as in Schloesing's second method. To this extract, magnesia is added and ozone is bubbled through the solu-

1. Analyse des Matieres Agricoles. 1879.
2. Agronomie 1864: 206.
3. Chem. Centralb. 1860: 243, 253.
4. Landw. Vers. Sta. 33: 247. 1886.
5. Jour. of Ag. Sci. 1910: 233.

tion, and it is then treated with sodium hypobromite and the nitrogen collected and measured. In view of the facts which are brought out below in regard to the hydrochloric acid extract of soils, together with the fact that the sodium hypobromite used contained an excess of sodium hydroxide, which would gradually decompose nitrogenous organic compounds to give ammonia, methods IV and V may be considered as unreliable.

VI. In Russel's first method, 150 grams of soil are distilled at the pressure obtained with a water pump with 2 grams of magnesia suspended in 100 c.c. of water. The distillation ask is kept at 40° C., and the distillation is continued for six hours.

VII. Russel's second method is similar to his first method, except that the 2 grams of magnesia are replaced by 0.7 gram potassium magnesia hydroxide and the 100 c.c. of water by an equal volume of alcohol, and the temperature of the distillation ask is kept at 25° C. instead of 40° C. Russel states that insofar as he has tested his methods, they give practically identical results. He also points out that with the use of magnesia there is some decomposition. Indeed, with the soil upon which he reports results he obtained both on the second and third distillation as much ammonia as was obtained on the first.

Although Russel shows that his potash method gives concordant results and no ammonia is given off after a reasonable length of time (first requirement), yet upon addition of ammonia to the soil, only from 50 to 84 per cent is recovered by either method. Hence, they do not comply with the second requirement. He explains this by stating that the ammonia enters into a stable combination which is not an ammonium compound. It could be argued with equal force that the ammonia forms a relatively insoluble ammonium compound with the complex organic or inorganic material of the soil, or that it was physically absorbed. It does not seem surprising that all the ammonia was not expelled when there was 50 c.c. of water left with the soil after the distillation was stopped, and the high solubility of ammonia[1] and the low solubility of magnesium hydroxide[2] is considered.

EXPERIMENTAL.

In the following work, the ammonia free water which was used was prepared by boiling ordinary tap water for a

1. Seidell, Solubilities of Inorganic and Organic Substances, 1911: 17. 30.7 grams per 100 grams water at 40° C. and 760 mm. pressure. At the pressure Russel used, the ammonia would be soluble to the extent of approximately one gram per 100 grams water.
2. Ibid.: 1. 181. 0.008-0.009 grams $Mg(OH)_2$ per liter of water at 18° C.

few minutes with about two grams of potassium acid sulphate to one liter of water, and then distilling the solution and condensing the vapor in block tin tubes which had previously been steamed for 12 hours. The distillate in every case was found to be neutral to alizarin red and gave but a slight indication of ammonia when tested with Nessler's solution.

Whenever magnesia was used, it was always freshly calcined.

Alizarin red was the indicator used in all ammonia titrations. This has been found to be by far the most satisfactory indicator for ammonia titrations, and is now exclusively used for that purpose in this laboratory.

Blanks were always run and the suitable corrections made. Duplicates were not run, except as reported.

The following soils were used in this work:

Soil No. 1—Southern Iowa loess.
Soil No. 2—Iowa drift.
Soil No. 3—Wisconsin drift.
Soil No. 4—Mississipi lopess.
Soil No. 5—Missouri loess.
Soil No. 6—Alkali I.
Soil No. 7—Alkali II.

Soils nos. 1 to 5 were typical samples from the five principal soil areas of Iowa. Soils nos. 6 and 7 were from the Wisconsin drift area.

AMMONIA IN THE HYDROCHLORIC ACID EXTRACT OF SOILS.

To find out whether the Schloesing method would give reliable results, it was examined as follows:

1. The strength of the hydrochloric acid used for extraction was varied.
2. The time of extraction was varied.
3. Known amounts of ammonia as sulphate were added to the soil and it was then extracted with different strengths of hydrochloric acid for different lengths of time.

All of the acid extracts were obtained by shaking in a mechanical shaking machine for the indicated length of time a mixture of one part of air dry soil with two parts of ammonia free acid. When ammonia was added, it was introduced as standard ammonium sulphate solution immediately before the addition of acid of suitable strength to provide the desired normality. Thus, any appreciable bacterial change of the ammonia was precluded. After removing from the shaking machine, the mixture was filtered through a double folded filter, a clear filtrate being obtained in all cases. For the magnesia distillation, 200 c.c. of the filtrate, repre-

senting 100 grame of soil, were distilled after the addition of about 10 grams of magnesia until 150 c.c. of distillate had been collected. In some cases 0.1 N acid and alkali were used in the titrations, and in others 0.02 N. The results of the magnesia distillations are given in table I.

TABLE I.

Soil No.	Lab. No.	Strength of Acid	Time Shaken, in Min.	Ammonia Added in Mgs. N	Ammonia Found in Mgs. N	Average in Mgs. N	Recovered in Mgs. N	Per Cent Recovered
1	1	0.2N	20 min.		1.26			
1	1	0.2N	"		1.19	1.23		
1	3	0.3N	"		1.26			
1	4	0.3N	"		1.26	1.26		
1	5	0.2N	60 min.		1.26			
1	6	0.2N	"		1.23	1.25		
1	7	0.3N	"		1.26			
1	8	0.3N	"		1.26	1.26		
1	9	0.2N	30 min.	2.25	2.60			
1	10	0.2N	"	2.25	2.60	2.60	1.35*	60.0
1	11	0.3N	"	2.25	2.60			
1	12	0.3N	"	2.25	2.64	2.62	1.37*	60.9
3	13	0.2N	20 min.		0.772			
3	14	0.3N	"		0.842			
3	15	0.2N	60 min.		0.814			
3	16	0.3N	"		0.842			
3	17	0.2N	20 min.	2.25	2.32		1.55	68.0
3	18	0.3N	"	2.25			1.38	61.3
3	19	0.2N	60 min.	2.25	2.25		1.44	64.0
3	20	0.3N	"	2.25	2.36		1.32	67.6

*In computing these values the average of the averages of nos. 1 to 8 were used

The results in table I show that within reasonable limits no ammonia enters into solution after 20 minutes, and that also within the limits of the experiment the amount of ammonia dissolved is independent of the strength of acid. It is also shown that not nearly all the ammonia which was added was recovered. The rather wide variance of the percentages recovered in the case of soil no. 3 is due to the error in the determination of so small an amount of ammonia as was in this soil.

Since the hydrochloric acid extract undoubtedly contains some organic nitrogenous material, it was of some interest to subject the residues in several of the distillation flasks to a second distillation. Accordingly, 150 c.c. of ammonia free water was added to each of the flasks of nos. 1 to 4 and 150 c.c. distilled as before. The results are given in table II.

It is seen that an appreciable amount of ammonia is given in each instance, and this no doubt comes from a decomposition of organic material. To further test this point

TABLE II.

Lab. No.	Residues from No.	Ammonia Found, in Mgs. N
21	1	0.28
22	2	0.27
23	3	0.42
24	4	0.39

and to test the applicability of Folin's[1] method for ammonia to the hydrochloric acid extract of soils, 100 c.c. portions of the same filtrates of which 200 c.c. were used in experiments nos. 1 to 4 were aerated for four hours, after the addition of 4 grams of sodium carbonate, in an apparatus which will be described later. The results are set forth in table III, and are for 100 grams of soil. For purposes of comparison, the results obtained by distillation with magnesia are repeated here.

TABLE III.

Lab. No.	From same Filtrate as was used in Nos.	Ammonia found by Aeration, in Mgs. N	Ammound found by Distillation, in Mgs. N
25	1 and 2	1.26	1.23
26	3 and 4	1.26	1.26
27	5 and 6	1.26	1.25
28	7 and 8	1.29	1.26

The aeration was continued for one more hour, but no ammonia was given. The results obtained by the aeration method do not vary from those found by distillation by more than the experimental error. Owing to the possibility of decomposition with magnesia at 100° C. one would expect the results obtained by aeration to be slightly lower. The results show that this method can be applied to the hydrochloric acid extract and its use in preference to the distillation method is recommended, both from the standpoint of accuracy and ease of manipulation.

In table IV are given the results obtained by aerating the acid filtrates from soils nos. 2, 4 and 5 as outlined above. For purposes of comparison, the results are calculated for 100 grams of soil, although as above they were obtained on 100 c.c. of filtrate, representing 50 grams of soil.

The results recorded in table IV bring out about the same points as those in table I, namely, that the amount of ammonia extracted is, within the limits of the experiment, independent of the strength of acid or the time of extraction, and that not all of the added ammonia is recovered. For the soils tested, and these represented quite a range of soil types, we are forced to the conclusion that any method which

1. Zeit. f. physiol. Chem. 37: 161. 1902.

TABLE IV.

Soil No.	Lab. No.	Strength of Acid	Time Shaken in Min.	Ammonia Added, in Mgs. N	Ammonia found in Mgs. N	Ammonia recovered in Mgs. N	Per Cent recovered
5	29	0.2N	20 min.		1.10*		
5	30	0.3N	"		1.21		
5	31	0.2N	60 min.		1.24		
5	32	0.3N	"		1.24		
5	33	0.2N	20 min.	2.25	2.63	1.40	62.2†
5	34	0.3N	"	2.25	2.63	1.40	62.2†
5	35	0.2N	60 min.	2.25	2.59	1.36	60.4†
5	36	0.3N	"	2.25	2.63	1.40	62.2†
2	37	0.2N	30 min.		1.24		
2	38	0.2N	"	2.25	2.80	1.56	69.3
4	39	0.2N	"		0.53		
4	40	0.2N	"	2.25	1.93	1.40	62.2

*Probably an error.
†In computing these results, the average of the results obtained in experiments 30, 31 and 32 was used.

requires an examination of the hydrochloric acid extract is not reliable. It might indeed be contended that treatment with fresh portions of hydrochloric acid would finally recover all the added ammonia. It is questionable whether this point could easily be proven, for no doubt long continued treatment of the soil even with acid of this dilution would gradually decompose organic matter with the production of amomnia. It was not thought worth while to investigate this point, for even if a method could be elaborated, it probably would not comply with the third requirement, namely that of practicability. As to the cause of the failure to recover all the added ammonia, any explanation would be purely speculative. In the course of some work which we shall publish later, we have found that the use of too rigorous flocculating agents to clear the solution for the determination of nitrates in the soil causes a decided lowering in the amounts of nitrates found. It is possible that the phenomenon of incomplete recovery of ammonia is due to the flocculating action of the acid.

DISTILLATION OF SOIL AT ORDINARY PRESSURE WITH MAGNESIA.

It has been shown[1] that successive distillation with magnesia of such substances as beef, eggs, dried blood and cottonseed meal gives for several distillations small but appreciable amounts of ammonia, but so far as we know, this method has never been critically examined with regard to

1. Trescott, Bul. U. S. Dept. of Ag., Bur. of Chem., 132: 20.

soil. We have accordingly carried out the following tests:

One hundred grams of soil were placed in a copper flask, together with 200 c.c. of ammonia free water, a small piece of paraffin and about 100 grams of magnesia and distilled until 150 c.c. of distillate had been collected in standard acid. The receiving flask was removed and replaced by another with a suitable quantity of standard acid. One hundred and fifty c.c. of ammonia free water were then put in the copper flask and 150 c.c. of distillate were again collected. This was repeated a third, and in the case of one soil, a fourth time. The results are given in table V.

TABLE V.

Soil No.	Lab. No.	Ammonia, 1st distillation, in Mgs. N	Ammonia, 2nd distillation, in Mgs. N	Ammonia, 3d distillation, in Mgs. N	Ammonia, 4th distillation, in Mgs. N
1	41	3.51	1.54	.91	
1	42	3.16	1.54	.91	
2	43	3.79	1.26	.84	
2	44	3.89	1.26	.84	
3	45	2.67	1.12	.81	
3	46	2.67	1.12	.84	
4	47	2.46	.98	.70	.70
4	48	2.53	.84	.70	.63
5	49	3.16	.77	.98	
5	50	3.26	.77	.91	

From the results recorded in table V, it is apparent that the amount of ammonia obtained by distillation is dependent upon the durtaion of the distillation, which is, of course, proportionate to the amount of water and soil used and upon the quantity of heat applied. The results of the following experiments emphasize the latter point in a striking way. For each experiemnt, 100 grams of soil, 200 c.c. of ammonia free water, a small piece of paraffin and 10 grams of magnesia were used. The full flame of a large burner was used on each of the flasks of nos. 51, 52, 55 and 56. The flames played directly upon the flasks, while in nos. 53, 54, 57 and 58, a low flame was used, and each flask was protected with a wire gauze. The results are given in table VI.

From a consideration of the results in table VI, although the conditions have been made extreme, there seems to be room for doubt as to whether even comparable results can be obtained by distillation with magnesia. Although one might regulate the gas flame during the distillation of a single series so that each gave about the same heat, yet the results obtained on different days or at a different time on the same day might be appreciably variable.

TABLE VI.

Soil No.	Lab. No.	Time in minutes of distillation	Mgs. N. as Ammonia
1	51	40	2.80
1	52	40	2.67
1	53	150	4.63
1	54	150	4.63
2	55	45	3.37
2	56	40	3.09
2	57	140	4.49
2	58	160	5.34

It is obvious, therefore, from the data presented above and from the work of others previously cited, that there is no very reliable method in use for the determination of ammonia in soils. That it is highly desirable to have such a method is apparent from the general importance of the soil nitrogen problem, particularly in the study[1] of the influence of lime and various other substances added to soil upon its ammonia content; in the study of the ammonia exaporation from soils; in the study[2] of the direct assimilation of ammonia by higher plants, and in the study of the ammonia problem in many other practical and theoretical phases. The attempt was made, therefore, to find a method for the determintaion of ammonia in the soil which would comply with the three requirements named in the introduction to this paper. The results as given below will show to what extent the attempt has succeeded.

AMMONIA IN THE SOIL BY AERATION.

In 1902, Folin[3] outlined the method which, with some modifications, is largely used for the determination of ammonia in urine and various other physiological products. Originally the method involved drawing air at the rate of 600 to 700 liters per hour for 1 to 1½ hours through 25 c.c. of urine containing 1 gram of sodium carbonate and 8 to 10 grams of sodium chloride. From the cylinder containing the urine the air passed through standard acid, which absorbed the ammonia. Steel and Gies[4] found that with urines containing magnesium ammonium phosphate crystals, low results were given because this salt was but slowly decomposed by sodium carbonate. It was later shown[5] that all urines,

1. Ehrenburg. Landw. Ztg. 60: 441, 479. 1911. Von Klodeck. Jour. Chem. Soc. (Eng.) II 102: 85; O. Lemmermann and L. Fresenius. Landw. Jahrb. 45: 127. 1913.
2. Hutchinson & Miller. Jour. Ag. Sci. III. 179.
3. Zeit. fur physiol. Chem. 37: 161. 1902.
4. Jour. Biol. Chem. 5: 71. 1909.
5. Benedict & Osterburg. Biecham. Bul. III, 41. 1913.

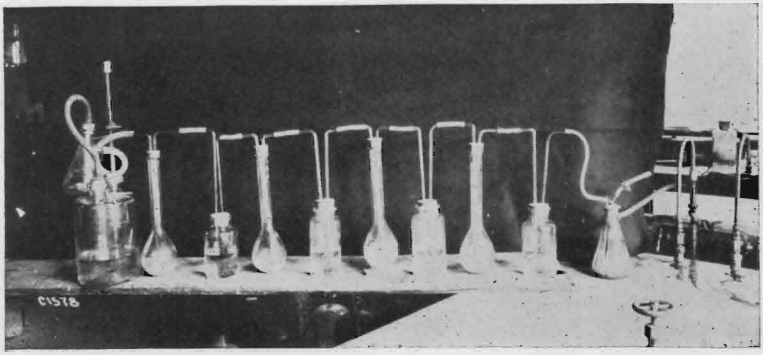
upon the addition of sodium carbonate, gave copious deposits of magnesium ammoniu mphosphate and, consequently, low results for ammonia. Steel[1], therefore, recommends the use of 0.5 gram sodium hydroxide in place of the 1 gram of sodium carbonate. He found that with the use of these reagents (sodium hydroxide and sodium chloride) all the ammonia was readily given off from triple phosphate crystals, and that apparently they did not decompose any organic materials, as no ammonia was given after four hours' aeration. He also found that aeration of solutions of each of the following substances gave no ammonia: Glycine, urea, uric acid, leucine, tyrosine, hippuric acid, guanine, allantoin, creatine and creatinine. Of these compounds, creatinine[2] and guanine[3] have been isolated from the soil by such methods as would make it appear that they were present as such and not in combination. Tyrosine[4] and leucine[5] have been isolated from the solution obtained by long continued boiling of the soil with hydrochloric acid.

Since the aeration methods have been so successfully used in connection with ammonia determinations in urine, which contains a wide variety of organic nitrogenous products, we decided to make an attempt to modify it for use with soil. Since ammonia has been shown by Russel[6] and the authors of this bulletin to be rather tenaciously held by the soil, the stronger reagent used in Steel's modification was first tried. In the course of some preliminary investigations, various forms of aeration aparatus were tried, but the form which was found most satisfactory was similar to the apparatus used by Kober[7] in his "Ammonia Distillation by Aeration" method. We use a 16 oz. bottle for the absorption bottle, and a 500 c.c. round bottom Kjeldahl flask for the aeration flask. The air and ammonia enter the absorption bottle through a specially made absorption tube. The directions for making this tube are given by Folin[8].

For any further details as to the setting up of the apparatus, the accompanying illustration should be consulted.

The filtration flask must always be set perpendicular to the table, and the long tube in the aeration flask should reach to within not more than 3 millimeters of the bottom of the flask. Both of these last precautions are necessary to secure adequate stirring of the mixture. Only 4 units are

1. Jour. Biol. Chem. 8: 365. 1910.
2. Sharey. Bul. U. S. D. of Ag. Bur. of Soils 83. 1911.
3. Lathrop. Jour. Am. Chem. Soc. 34: 1260. 1912.
4. Suzuki. Bul. Coll. Tokyo 7: 513.
5. Robinson. Tech. Bul. Mich. Ag. Exp. Sta. 7.
6. Loc. Cit.
7. Jour. Am. Chem. Soc. 35: 1594. 1913.
8. Loc. Cit.



The Aeration Apparatus

shown in the cut, but as many as 14 determinations in series on one pump have been run. For all the work reported in this paper, a current of air of about 250 liters per hour was used.

In our trial of the Steel method, the following technique was used: The "alkali" was prepared by saturating ammonia free water wtih sodium chloride. In order to be always sure that a saturated solution was obtained, the calculated amount of the salt was added to a known volume of water. Sodium hydroxide was added at the rate of 2 grams per 100 c.c. of water. Twenty-five grams of soil were placed in a Kjeldahl flask. The absorption bottle was half filled with ammonia free water[1] and 100 c.c. of 0.02 N. sulphuric acid added. The apparatus was then connected up, the stopper replaced and the flask shaken and the suction started.

The results are set forth in table VII, and are computed to 100 grams of air dry soil. For purposes of comparison, the results obtained for the corresponding soils on the first distillation with magnesia are included.

Since after aerating 15 hours ammonia was being expelled in relatively large amounts, and because the amounts given off are far in excess of those given off by distillation with magnesia, there is no other conclusion but that there is a decided decomposition of organic nitrogenous matter to give ammonia. The work of Steel[2] shows that probably this excess ammonia is not due to a decomposition of amino acids. Another class of nitrogenous compounds less stable

1. The reason for using ammonia free water here was that a sharper end point was obtained with it than when ordinary distilled water was used in the nitration. This was, no doubt, due to a slight trace of amphoteric matter in the distilled water.
2. Loc. Cit.

TABLE VII.

Soil No.	Lab. No.	Time of aeration in hours	Ammonia in Mgs. N	Ammonia by MgO, in Mgs. N
1	59	4½	3.87	
1	60	4½	3.93	
1	61	6	4.49	
1	62	6	4.63	
1	63	8	5.61	
1	64	8	5.56	
1	65	15	7.02	
1	66	15	7.30	
1	67	19	8.70	
1	68	19	8.99	3.33
3	69	15	4.21	
3	70	15	4.49	
3	71	19	6.60	
3	72	19	6.46	2.67
5	73	15	7.58	
5	74	15	7.86	
5	75	19	9.13	3.21
5	76	19	9.41	

in the presence of alkalies than the amino acids is the acid amides. Kelly and Thompson[1] in analyses of the alkali extract of nine soils, found an average of 14.3 per cent amide nitrogen. Jodidi[2], Robinson[3] and others have found from 20 to 30 per cent of amide nitrogen in the solution obtained by boiling the soil for several hours with strong acid. But because of their inherent instability, it seems highly improbable that acid amides are present in soils in much larger, if as large, percentages as ammonia. Many acid amides would slowly decompose in the presence of the Steel reagent. For instance, we found that 0.1 gram acetamide, purified by recrystallization from ether, in two hours aeration with the Steel reagent, gave 1.54 mgs. ammonia and in four hours, 2.56 mgs. It seem squite probable, therefore, that at least part of the excess ammonia is due to the decomposition of acid amides.

It was next decided to try the use of sodium carbonate as the alkali. Accordingly, as a preliminary test, 0.1 gram of acetamide was dissolved in 50 c.c. of water, 2 grams of sodium carbonate added and the solution aerate. At the end of two hours, 0.08 mg., and at the end of twenty hours 0.02 mg. of ammonia was given. No doubt the production of even the 0.08 mg. during the first two hours was from a trace of ammonium salts in the acetamide.

1. Jour. Am. Chem. Soc. **36**: 438. 1914.
2. Tech. Bul. Mich. Ag. Exp. Sta. 4. 1909. Research Bul. Iowa Ag. Exp. Sta. 1 and 3. 1911.
3. Loc. Cit.

As is stated above, it has been found that all urines upon the addition of sodium carbonate give triple phosphate crystals, and on aeration low results are given for ammonia. Since magnesium, ammonium and phosphate are present in the soil solution, it would seem that the same thing would take place as with urines, namely, low results with sodium carbonate. In 1908 Folin[1] found that this objection in the case of urine could be overcome by the addition of potassium oxalate to the solution before the addition of sodium carbonate. If the magnesium ammonium phosphate was already there as a precipitate, the solution should first be made acid, which would dissolve the crystals and then the potassium oxalate added. This same procedure could, of course, be applied directly to any solution besides urine. We have, therefore, tried the use of sodium carbonate with and without the addition of potassium oxalate on seven soils, and in the case of one soil, we have tried sodium chloride and sodium carbonate. The results are all grouped together in table VIII, and are for 100 grams of soil.

In all cases, 25 grams of soil, 50 c.c. of ammonia free water and about 2 grams of sodium carbonate, and where indicated 8 to 10 grams of potassium oxalate and 15 to 17 grams of sodium chloride, were used. Also, when potassium oxalate was used, the mixture was always made slightly acid before the addition of the sodium carbonate.

An examination of the foregoing table brings out many interesting points. For the soils tested, sodium carbonate used alone causes no appreciable decomposition of organic matter. It is possible that with a soil high in organic matter, it might be advantageous to use sodium chloride, which, of course, acts to weaken the base. The use of potassium oxalate, it is seen, gives no more ammonia than sodium carbonate alone. This is at first thought somewhat surprising, but no doubt the reason for this is that the phosphate and ammonia are present in such high dilution in the soil solution that the magnesium ammonium phosphate does not precipitate. In urine[2] the phosphate content averages about 2000 parts per million, and the ammonia[3] 1000 parts per million, while in the soil King[4] found as a maximum amount of water soluble phosphate about 20 parts per million, and we have found as a maximum amount of ammonia about 30 parts per million. Since by our method 2 parts of water to 1 part of soil are used, these values should be halved. It is,

1. Jour. Biol. Chem. 8: 497. 1910.
2. Hammarsten. Text Book of Physiological Chemistry. 646. (6th Eng. ed.)
3. Hawk's Practical Physiological Chemistry. 313. (4th ed.)
4. King. Bul. U. S. Dept. of Ag., Bur. of Soils. 26.

TABLE VIII.

Soil No	Lab. No.	Time of aeration in hours	Alkali reagent	Ammonia added, in Mgs. N	Ammonia found in Mgs. N	Average, in Mgs. N	Ammonia recovered in Mgs. N	Per Cent recovered
1	78	15	Na^2CO^3, NaCl		2.08			
1	79	15	" "		2.08	2.08		
1	80	15	" "	2.25	4.29			
1	81	15	" "	2.25	lost	4.29	2.21	98.2
1	82	19	" "		1.97			
1	83	19	" "		2.08	2.03		
1	84	19	" "	2.25	4.29			
1	85	19	" "	2.25	4.27	4.28	2.25	100
1	86	19	Na_2CO_3		2.08			
1	87	19	"		1.97	2.03		
1	88	19	"	2.25	4.27			
1	89	19	"	2.25	4.32	4.30	2.27	100.9
1	90	15	$K_2C_2O_4$, Na_2CO_3		2.08			
1	91	15	" "		2.08	2.08		
1	92	15	" "	2.25	4.32			
1	93	15	" "	2.25	4.27	4.30	2.22	98.7
1	94	19	" "	2.25	4.32			
1	95	19	" "	2.25	4.32	4.32	2.24	99.5
2	96	15	Na_2CO_3		2.59			
2	97	15	"		2.53	2.56		
2	100	15	"	2.25	4.84			
2	101	15	"	2.25	4.78	4.81	2.25	100
2	102	19	"		2.53			
2	103	19	"		2.55	2.54		
2	104	19	"	2.25	4.60			
2	105	19	"	2.25	4.78	4.78	2.24	99.5
2	106	15	$K_2C_2O_4$, Na_2CO_3		2.59			
2	107	15	" "		2.59	2.59		
2	108	15	" "	2.25	4.84			
2	109	15	" "	2.25	4.78	4.81	2.22	98.7
2	110	19	" "	2.25	4.71			
2	111	19	" "	2.25	7.84	4.78	2.19	97.3
3	112	15	Na_2CO_3		1.23			
3	113	15	"		1.23	1.23		
3	114	15	"	2.25	3.48			
3	115	15	"	2.25	3.54	3.51	2.28	101.3
3	116	19	"		1.23			
3	117	19	"		1.18	1.21		
3	118	19	"	2.25	3.48			
3	119	19	"	2.25	3.48	3.48	2.27	100.9
3	120	15	$K_2C_2O_4$, Na_2CO_3		1.23			
3	121	15	" "		1.18	1.21		
3	122	15	" "	2.25	3.48			
3	123	15	" "	2.25	3.54	3.51	2.30	102.1
3	124	19	" "	2.25	3.48			
3	125	19	" "	2.25	3.48	3.48	2.27	100.9
4	126	15	Na_2CO_3		1.39			
4	127	15	"		1.39	1.39		
4	128	15	"	2.25	3.59			
4	129	15	"	2.25	3.65	3.62	2.23	99.1
4	130	19	"		1.29			
4	131	19	"		1.34	1.32		
4	132	19	"	2.25	3.59			
4	133	19	"	2.25	3.59	3.59	2.27	100.9
4	134	15	$K_2C_2O_4$, Na_2CO_3		1.34			
4	135	15	" "		1.34	1.34		
4	136	15	" "	2.25	3.59			
4	137	15	" "	2.25	3.59	3.59	2.25	100
4	138	19	" "	2.25	3.54			
4	139	19	" "	2.25	3.59	3.57	2.23	99.1
5	140	15	Na_2CO_3		2.35			
5	141	15	"		2.19	2.27		
5	142	15	"	2.25	4.44			
5	143	15	"	2.25	4.49	4.47	2.20	97.8
5	144	19	"		2.25			
5	145	19	"		2.25	2.25		

5	146	19	"		2.25	4.44			
5	147	19	"		2.25	4.44	4.44	2.19	97.3
5	148	15	$K_2C_2O_4$,	Na_2CO_3		2.19			
5	149	15	"	"		2.25	2.23		
5	150	15	"	"	2.25	4.44			
5	151	15	"	"	2.25	4.49	4.47	2.24	99.5
5	152	19	"	"	2.25	4.49			
5	153	19	"	"	2.25	4.49	4.49	2.26	100.5
5	154	15	Na_2CO_3			0.79			
6	155	15	"			0.79	0.79		
6	156	15	"		2.25	3.03			
6	157	15	"		2.25	3.03	3.03	2.24	99.5
6	158	19	"			0.79			
6	159	19	"			0.84	0.82		
6	160	19	"		2.25	3.03			
6	161	19	"		2.25	3.14	3.09	2.25	100
6	162	15	$K_2C_2O_4$,	Na_2CO_3		0.90			
6	163	15	"	"		0.79	0.85		
6	164	15	"	"	2.25	3.03			
6	165	15	"	"	2.25	3.09	3.06	2.21	98.2
6	166	19	"	"	2.25	3.09			
6	167	19	"	"	2.25	3.09	3.09	2.24	99.5
7	168	15	Na_2CO_3			1.12			
7	169	15	"			1.12	1.12		
7	170	15	"		2.25	3.37			
7	171	15	"		2.25	3.26	3.32	2.20	97.8
7	172	19	"			1.23			
7	173	19	"			1.18	1.21		
7	174	19	"		2.25	3.48			
7	175	19	"		2.25	3.37	3.43	2.22	98.7
7	176	15	$K_2C_2O_4$,	Na_2CO_3		1.12			
7	177	15	"	"		1.23	1.18		
7	178	15	"	"	2.25	3.37			
7	179	15	"	"	2.25	3.37	3.37	2.19	97.3
7	180	19	"	"	2.25	3.37			
7	181	19	"	"	2.25	3.48	3.43	2.25	100.0

therefore, apparent why there is no interference of the triple phosphate.

From the data presented in table VIII, it is safe to recommend the aeration method with sodium carbonate as the alkali for ammonia determination in normal and Iowa alkali soils. The method complies with the three requirements laid down as fundamental. Closely concordant duplicates are obtained, no appreciable amount of ammonia is given off after fifteen hours aeration, and with the experimental error 100 per cent of any added ammonia was always recovered. The method is eminently practical.

For purposs of comparison, the results obtained by the three methods are assembled in table IX.

TABLE IX.

Soil No.	Ammonia HCl Extraction in Mgs. N	Ammonia MgO distillation in Mgs. N	Ammonia Na_2CO_3 aeration in Mgs. N
1	1.25	3.33	2.03
2	1.24	3.84	2.56
3	0.80	2.67	1.23
4	0.53	2.50	1.39
5	1.23	3.21	2.27

It is seen that there is very little correlation in the amounts of ammonia found by the three methods, except that extraction with the hydrochloric acid gives lower re-

sults, while distillation with magnesia gives higher results than the aeration method. This is to be expected, since we have shown that hydrochloric acid holds back ammonia, while distillation with magnesia effects some decomposition. Contrary to our findings, Kelley and McGeorge[1] report concerning the ammonia determinations in the hydrochloric acid extract, that the "results were very similar to those obtained by direct [magnesia] distillation." Table IX emphasizes the fact that in reporting ammonia determinations in soil, the method used should in all cases be stated.

For the convenience of those who wish to use the aeration method for ammonia in soils, the technique which has been used will be given here in detail. After all the apparatus is at hand, the procedure is as follows: Prepare the absorption bottles as was described above for the Steel emthod. Weigh 25 grams of soil into the 500 c.c. Kjeldahl flasks; add 50 c.c. of ammonia free water and a few drops of a heavy oil to prevent foaming, and then insert the rubber stopper bearing the tube for the entrance and exit of the air. Have the end of the long glass tube come within not more than 3 millimeters of the bottom of the flask. See that no water is between the rubber stopper and the mouth of the flask to collect ammonia. The best way to eliminate this danger is to have the rubber stopper fit very tightly. Now, connect up the whole series, but do not start the pump. As many as fourteen determinations can be run in series. The air, before entering the system, must, of course, be passed through a wash bottle containing dilute sulphuric acid. Now loosen all the rubber stoppers in the Kjeldahl flasks, start the pump, add about 2 grams of sodium carbonate to the flask nearest the pump, shake the flask and then insert the rubber stopper and then take the flasks in succession in just the same way. After a series is once started, the pump should not be stopped. If anything happens to a determination, do not try to remove it. After the aeration has run as long as desired, with the pump still going remove the flasks one by one, starting with the one farthest removed from the pump.

This same apparatus has been used by us for the determination of total nitrogen in soil. The digestion is carried out in the usual way, and the aeration conducted in much the same way as Kober[2] recommends. A water bath, as he advises, was not used, but instead, add about one-third of the required amount of alkali; shake and allow the mixture to cool and then add the remainder of the alkali by the same

1. Bul. Hawaiian Sta. 30: 31. 1913.
2. Loc. Cit.

procedure he uses, except the absence of the water bath. To prevent spattering, the Kjeldahl flask is tilted in a direction perpendicular to a line connecting the two adjacent absorption bottles. As to accuracy and ease of manipulation, we can confirm Kober's statements.

As has been stated, all aeration work reported in this paper is for a current of air of about 250 liters per hour. If a pump is available which moves more air than this, no doubt the time of aeration could be correspondingly lessened.

CONCLUSION.

1. The amount of ammonia extracted by hydrochloric acid is within the limits of our experiments, independent of the strength of the acid and the period of extraction.
2. In the five soils tested, hydrochloric acid removes approximately from 60 to 70 per cent of the ammonia added.
3. The Folin aeration method can advantageously be applied directly to the hydrochloric acid extract.
4. The amounts of ammonia obtained by distillation of the soil directly with magnesia is dependent upon the duration of the distillation.
5. The Steel method of aeration is not suitable for the determination of amomnia in soils.
6. The Steel reagents slowly decompose acetamide.
7. The Folin method of aeration is suitable for the determination of ammonia in soils, for the same result is obtained, whether the reagent acts for a shorter or longer period, and all added ammonia is recovered.
8. In the soils tested there is no interference through formation of triple phosphate.
9. For the soils tested, there was no advantage found in using sodium chloride with the sodium carbonate.
10. Acetamide is not decomposed by 4 per cent sodium carbonate.
11. The results for ammonia obtained by examination of the hydrochloric acid extract are lower, while the results obtained by direct distillation of the soil with magnesia are higher than those obtained by the aeration method. The high results obtained by the former method are due to occlusion of the ammonia y the soil, the nature of which is not clear, and the lower results by the latter method are due to a partial decomposition of the organic material by the magnesia to give ammonia.